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PRONIOSOMAL GEL: A NOVEL VESICULAR APPROACH FOR CONTROLLED DELIVERY OF NIFEDIPINE

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ABSTRACT

Nifedipine is a calcium channel blocker extensively used in the treatment of anginal and hypertension. On oral administration it undergoes extensive first pass metabolism, which outweighs its absorbance through gastrointestinal tract (GIT) and bioavailability of the drug in systemic circulation. As an alternative to oral route transdermal route of drug delivery was developed. The formulated systems were characterized in vitro for size, vesicle count, drug entrapment and drug release profiles. In the present investigation, proniosomes are prepared by varying the ratio of span-20, span-60, tween20, Tween 60, lecithin, cholesterol, aqueous phase and polymer. Formulation containing Tween20 and Tween 60, lecithin, Cholesterol, alcohol, 0.1% glycerol (800:100) showed optimal vesicle size, high entrapment efficiency. The Franz-diffusion studies were carried out in pH 7.4 using cellophane membrane and the results showed better permeability of niosomes with good steady state flux and enhancement ratio suggesting the potential of proniosomal carriers for improved transdermal delivery of nifedipine.

Key Words: Nifedipine, Proniosomes, Transdermal, Permeability,Niosomes

INTRODUCTION:

The Novel drug delivery system should preferably fulfil two prerequisites which are; firstly, it should deliver the drug at a rate directed by the needs of the body, till the period of treatment; Secondly, it should channel the active ingredient of the formulation to the site of action[1].

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In the past, the delivery of the drugs has been altered by routes such as topical delivery has been widely explored. The rapid development of transdermal delivery formulations in the last years is due to its ability to overcome certain problems of the conventional system of drug development. One of the systems to overcome the problems through transdermal route is vesicular approach. Proniosomes offer a versatile vesicle drug delivery concept with potential for delivery of drugs via transdermal route[2]. This would be possible if proniosomes form niosomes upon hydration with water from skin following topical application

under occlusive conditions[3]. Proniosomes minimizes problems of niosomes physical stability such as aggregation, fusion and leaking and provide additional convenience in transportation, storage and dosing[4]. Hypertension is an increasingly important medical and public health issue. Hypertension plays a major role in the development of cerebrovascular disease, ischemic heart disease, cardiac and renal failure. Several classes of medications, collectively referred to as antihypertensive drugs, are currently available for treating hypertension[5]. Nifedipine, a calcium channel blocker used in the treatment of hypertension and angina pectoris. Nifedipine (20–60 mg) once-daily, orally given in the treatment of hypertension. Its solubility is poor in both lipophilic and hydrophilic media. The treatment requires a constant release of the drug into systemic circulation. Since, its half life is 2-4 hrs requires frequent dosing of the drug. Even though Nifedipine is rapidly and almost completely absorbed from GI tract but it undergoes extensive first pass metabolism (around 60%) resulting in a poor bioavailability (45%) after oral administration. Hence, to improve its therapeutic efficacy, patient compliance and to reduce the frequency of dosing and side effects as well as to avoid its extensive first pass metabolism, transdermal drug delivery approach was considered to be better suitable for Nifedipine[6]. The various vesicular systems, as a transdermal carrier, have been gaining attention because they not only act as depot for delivery of contents but also act as penetration enhancers. The various carrier systems like liposomes, ethosomes and transfersomes comprising of phospholipid that are reported to be harmless and non irritating to the skin but the poor stability is the major problem in the development of these vesicular systems at industrial and clinical levels[7]. In order to overcome the stability problem these are characterized as proniosomes which converts into niosomes on hydration. Proniosomes provide higher stability and better skin penetration ability than the traditional lipid vesicles, e.g. liposomes, niosomes etc[8].

Materials and Methods

Nifedipine was provided by Hetero drugs Pvt ltd. Soya lecithin was purchased from Hi media- Bombay, Cholesterol was purchased from Hi media chemicals Bombay. All other chemicals used throughout this investigation were of analytical grade and no additional purification was carried out. Double distilled water was used throughout the study

Estimation of Nifedipine[9]:

Preparation of calibration curves

Nifedipine solution was scanned in the u.v range of 200 – 400 nm using systronic double beam u.v visible spectrophotometer.

Determination of wavelength of maximum absorbance (λ max)

10mg of drug was weighed accurately and transferred to 10ml of volumetric flask. Then phosphate buffer 7.4 (suitable solvent) was added to dissolve the drug completely. The volume was made up to 10ml with solvent. The prepared sample was 1000 µg/ml. 1ml of above solution was then transferred to another 100ml volumetric flask and diluted it up to the mark with phosphate buffer 7.4. This sample was 10µg/ml.

Preparation of calibration curve of Nifedipine

The calibration curve was plotted between the concentration and absorbance. The concentrations ranging from 1-25ug/ml were used to construct calibration curve. The results for calibration curve shown in table.1 and figure.1

Development of Proniosomal Gel

Proniosomal gel was prepared by a Coacervation-phase separation method. Precisely weighed amounts of surfactant, lecithin, cholesterol and drug were taken in a clean and dry wide mouthed glass vial of 5.0 ml capacity and alcohol (0.5 ml) was added to it. After warming, all the

ingredients were mixed well with a glass rod; the open end of the glass bottle was covered with a lid to prevent the loss of solvent from it and warmed over water bath at 60-70°C for about 5 min until the surfactant mixture was dissolved completely. Then the aqueous phase (0.1% glycerol solution) was added and warmed on a water bath till a clear solution was formed which was converted into proniosomal gel on cooling. The gel so obtained was preserved in the same glass bottle in dark conditions for characterization. Compositions of proniosomal gel formulations are given in Table 2[10].

Characterization of Proniosomal Gel

Vesicle Size Analysis:

Hydration of proniosomal gel (100mg) was done by adding saline solution (0.9% solution) in a small glass vial with occasional shaking for 10 min. The dispersion was observed under optical microscope at 100 x magnification. The sizes of 200-300 vesicles were measured using a calibrated ocular and stage micrometer fitted in the optical microscope[10].

Entrapment efficiency

To evaluate the loading capacity of proniosomal systems for Nifedipine, proniosomal gel (100mg) was dispersed in distilled water and warmed a little for the formation of niosomes. Then the dispersion was centrifuged at 18000 rpm for 40min at 5ºC. The clear fraction was used for the determination of free drug at 238.0 nm spectrophotometrically. The percentage encapsulation efficiency was calculated from Equation. % Encapsulation Efficiency = [1-(Unencapsulated drug / Total drug)] x 100

Rate of Spontaneity

Approximately 10 or 20 mg of proniosomal gel was transferred to the bottom of a clean stoppered glass bottle and spread uniformly around the wall of the glass bottle with the help of a glass rod. At room temperature, 2 ml of saline (0.9% NaCl) was added carefully along the walls of the glass bottle and left in a test-tube stand. After 20 minutes, a drop of this saline solution was withdrawn and placed on Neubauers Chamber to count the number of vesicles. The number of niosomes eluted from proniosomes was counted[11].

Surface morphological studies:

The surface morphology of noisome derived from proniosomal gel was studied using scanning electron microscopy. SEM revealed that the niosomes formed were spherical and homogeneous[12].

***In-Vitro* Release**

The *in vitro* drug release studies of proniosomal gel were carried out by means of treated cellophane membrane. In-vitro release studies on proniosomal gel were performed using Franz-diffusion cell. The capacity of receptor compartment was 10 ml[13,14]. The dialysis cellophane membrane was mounted between the donor and receptor compartment. A weighed amount of proniosomal gel was placed on one side of the membrane. The receptor medium was saline phosphate buffer pH 7.4. The receptor compartment was surrounded by a water jacket to maintain the temperature at 37±1ºC. Heat was provided using a thermostatic hot plate with a magnetic stirrer. The receptor fluid was stirred by a Teflon-coated magnetic bead fitted to a magnetic stirrerr[15]. At each sampling interval, (1 ml) were withdrawn and were replaced by equal volumes of fresh receptor fluid on each occasion. Samples withdrawn were analyzed spectrophotometrically at 238 nm[16].

***In vitro* permeation study:**

The permeation of Nifedipine from proniosomal formulations was determined by using Franz diffusion cell. The shaved abdominal skin of sheep (3.14 cm^2 exposed surface areas) was mounted on the receptor compartment with the stratum corneum side facing upwards towards the donor compartment. The receptor compartment was filled with 10.0 ml of pH 7.4 phosphate buffer maintained at 37.

8°C and stirred by a magnetic bar at 600 rpm. Weighed quantity of proniosomal gel formulation was placed on the skin and the top of the diffusion cell was covered with paraffin paper. At appropriate time intervals (3, 6, 9, 12, 18, 21, and 24 h), 1 ml aliquots of the receptor medium were withdrawn and immediately replaced by an equal volume of fresh receptor solution to maintain sink conditions Samples withdrawn were analyzed spectrophotometrically at 238 nm[17].

RESULTS AND DISCUSSION

Proniosome gel containing Nifedipine were prepared by Coacervation phase separation method. Formation of vesicle mainly depends on the concentration of cholesterol and surfactant ratio. Nifedipine solution was scanned in the UV range of 200-400nm using systronic u.v-visible spectrophotometer. The Spectrophotometric method of analysis of Nifedipine at λmax 238 nm was found to be reproducible and highly sensitive. The standard curves of Nifedipine were prepared in ethanol and phosphate buffer solution (pH 7.4), at λmax 238 nm. The data were regressed to obtain the straight line. The correlation coefficient greater than 0.99 was observed in all the cases, which indicated that, the drug follows Beer-Lambert's law in the concentration range of 10mg/10ml (Table 1 and Figure 1). Results of Vesicle size of Nifedipine proniosome are presented in (Table 3), which indicated that Vesicle formed with Span is smaller in size than vesicle formed with Tweens; this is due to grater hydrophobicity of Spans than Tweens. It is indicated that increasing in hydrophobicity decreases surface energy of surfactants resulting in smaller vesicle size. Size of vesicle was reduced when dispersion was agitated. The reason for this is the energy applied in agitation which results in breakage of larger vesicles to smaller vesicles. The size range was found to be 2.80±0.025µm to 7.45±2.10µm. Surface morphological studies revealed that proniosomes formed were spherical and homogeneous as shown in (Figure.2). For rate of spontaneity studies proniosomal formulation were treated with ethanol. It was found that proniosome with ethanol shows higher value as result of less phase separation which is due to their better solubility in water. Entrapment efficiency was found to be higher in case of proniosome prepared with Span20 and Span60 than proniosome prepared with Tween this is due to fact that Span 20 and Span 60 is more hydrophobic than Tween, which act as solid at room temperature and showed higher phase transition temperature (Tc), low HLB value and long alkyl chain length and results are shown in (Table 3). *In vitro* release studies are often performed to predict how a delivery system might work in an ideal situation as well as give some indications of its *in vivo* performance since drug release dictates the amount of drug available for absorption. The amount of drug released from different proniosomal gel formulation was found in the order of NG7 > NG8 > NG6 > NG5 > NG4 > NG3>NG2>NG1 as shown in (Figure.3). It was found that NG7 showed controlled release property from 10 to 24 hrs. The cumulative release found to 79.03% at the 24th hrs, respectively. The release rate was constant from 10thto 24th hrs. From kinetic release studies, it was confirmed that the release of drug follows zero order shown in Table.4. Thus the formulation exhibited zero order release over this period. *In vitro* permeation for optimized formulation through rat abdominal skin was found to be 73.645% after 24 hrs of release and data presented in (Figure.4) respectively. It was found that permeation of Nifedipine from proniosomal gel formulation prepared with Tween is slower as compared to proniosomal gel formulation prepared with span. This was expected due to the larger size of the vesicles and the less lipophilic nature of the former, which makes it more difficult for these vesicles to penetrate or fuse with the skin.

Table.1: Calibration Curve of Nifedipine

S.No.	Drug Conc. (µg/ml)	Absorbance (nm)
1	5	0.265
2	10	0.386
3	15	0.542
4	20	0.642
5	25	0.874

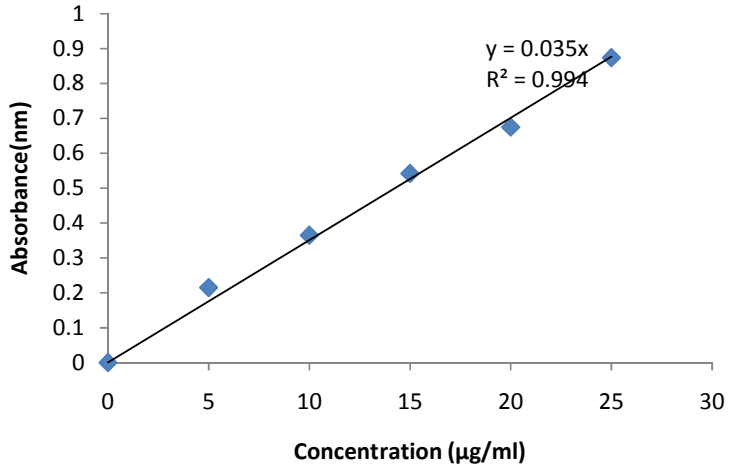

Figure.1: Calibration Curve of Nifedipine

Table.2: Composition of Nifedipine proniosomal gel formulations

Formulation code	Surfactant Type	Surfactant ratio	Cholesterol (mg)	Lecithin (mg)	Drug (mg)	Alcohol (ml)
NG1	Span20	900	100	100	20	0.5
NG2	Span60	900	100	100	20	0.5
NG3	S20:60	800:100	100	100	20	0.5
NG4	S20:60	100:800	100	100	20	0.5
NG5	Tween 20	900	100	100	20	0.5
NG6	Tween 60	900	100	100	20	0.5
NG7	T20:60	800:100	100	100	20	0.5
NG8	T20:60	100:800	100	100	20	0.5

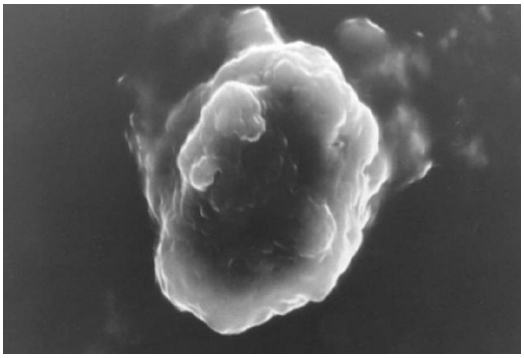

Figure 2: Scanning electron micrograph of optimized Nifedipine proniosomal gel formulation (NG7). (500X magnification)

Table.3: Entrapment Efficiency and Vesicle size of hydrated proniosomes

Formulation Code	Vesicle Size (µm) ±S.D.	% Entrapment Efficiency
NG1	4.26±0.24	66.69
NG2	3.61±0.15	71.23
NG3	2.80±0.025	74.35
NG4	3.36±0.196	76.27
NG5	5.26±0.46	77.89
NG6	6.26±0.12	78.42
NG7	6.56±0.15	80.43
NG8	7.45±0.15	78.35

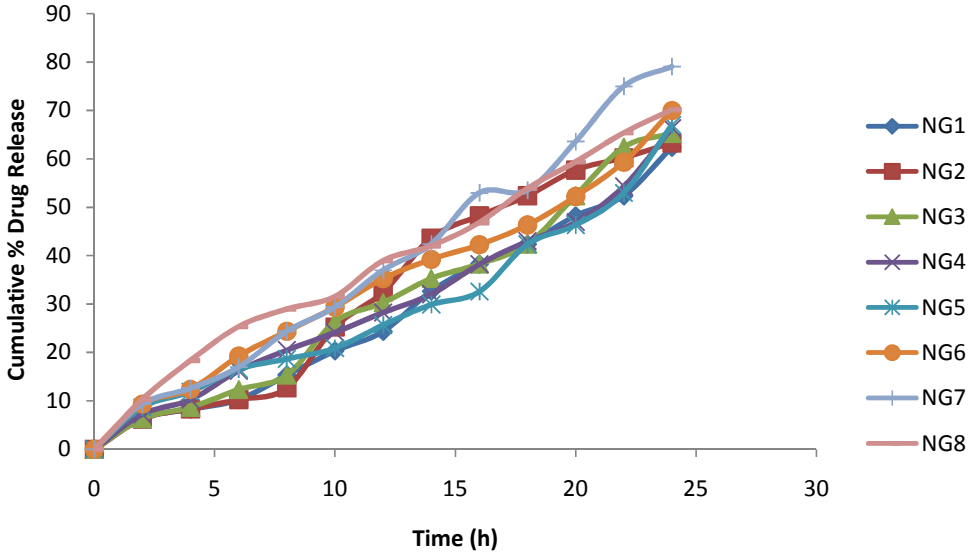

Figure.3: Comparison of *In vitro* Release studies for formulations NG1-NG8

Table.4: Kinetic Data for drug release from different formulations

Batch code	Regression for *In-vitro* plot (r^2)	Regression for Higuchi's plot (r^2)	Slope for Peppa's plot (n)
NG1	0.996	0.996	0.650
NG2	0.996	0.996	0.642
NG3	0.997	0.997	0.493
NG4	0.994	0.996	0.493
NG5	0.993	0.990	0.707
NG6	0.995	0.990	0.674
NG7	0.998	0.996	0.590
NG8	0.995	0.993	0.824

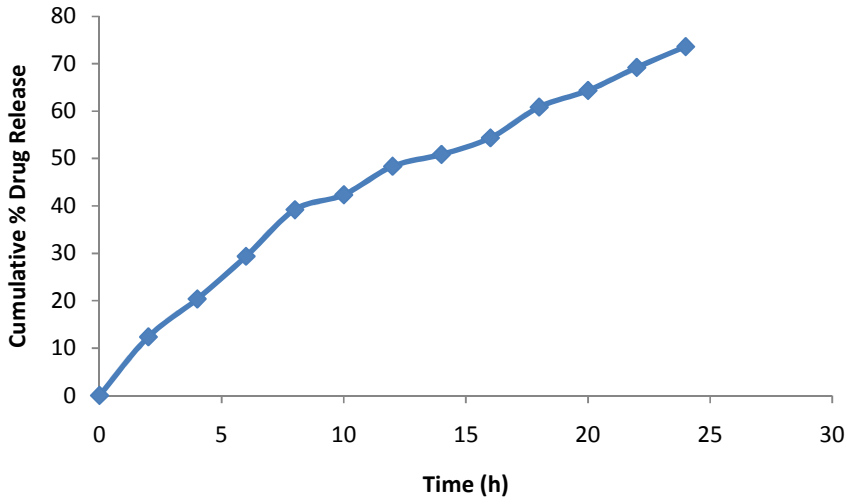

Figure.4: *In vitro* Permeation of drug from Formulation NG7

CONCLUSION:

The results of investigation revealed that proniosomes offers an alternative colloidal carrier approach in transdermal drug delivery. The results obtained from the present study clearly revealed that proniosomal gel containing Nifedipine which is prepared by using Coacervation phase separation method are capable of releasing drug for the extended period of time.

REFERENCES

1. Li Y & Mc Clements DJ. New mathematical model for interpreting pH-Stat digestion profiles: Impact of lipid droplet characteristics on in vitro digestibility. J. Agri and Food Chemistry, 2010; 58: 8085-8092.
2. Sudhamani T, Priyadarisini N, Radhakrishan M. Proniosomes– A Promising Drug Carrier. Int. J. Pharm. Tech. Res, 2010; 2(2): 1446-1454.
3. Fang JY, Yu SY, Wu PC, Huang YB, Tsai YH. *In-vitro* skin permeation of estradiol from various proniosomes formulation. Int. J. Pharm 2001; 215: 91-99.
4. Hu C, Rhodes DG. Proniosomes: A Novel Drug Carrier Preparation. Int. J. Pharm, 1999; 185(1): 23-35.
5. Turnbull F. Effects of different blood pressure lowering regimens on major cardiovascular events: results of prospectively- designed overviews of randomised trials. Lancet, 2003; 362(9395):1527-1535.
6. Dubey V, Mishra D, Nahar M, Jain NK. Vesicles as tools for the modulation of skin permeability. Expert Opin. Drug Deliv, 2007; 4(6): 579-593.
7. Gupta V, Agrawal RC, Trivedi P. Reduction in cisplatin genotoxicity (micronucleus formation) in non target cells of mice by protransfersome gel formulation used for management of cutaneous squamous cell carcinoma. Acta Pharm, 2011; 61(1): 63–71.
8. Jain S, Sapre R, Umamaheswari RB, Jain NK. Protransfersomes for effective transdermal delivery of norgestrel preparation and *in vitro* characterization. Indian J Pharm Sci, 2003; 65(2):152-160.
9. P. Neeraja, M. Amaleshwari and G. Ravali. Formulation and evaluation of multiple emulsion. Int. J. of

pharm, chemical and biological sci, 2014; 4(3): 673-680.
10. Ankur Gupta, Sunil Kumar Prajapati, M Balamurugan, Mamta Singh, Daksh Bhatia. Design and Development of a Proniosomal Transdermal Drug Delivery System for Captopril. Trop. J. Pharm. Res, 2007; 6 (2): 687-693
11. Jain NK, Vora B, Khopade AJ. Proniosome based transdermal delivery of levonorgestrel for effective contraception. J Control Rel,1998; 54: 149-165.
12. Prakash Goudanavar, Ashish Kute, , Doddayya Hiremath, S.R. Reddy. Development and Characterization of Perindopril Erbumine Loaded Proniosomal Gel. Asian J. Pharm. Tech. 2012; 2(2): 54-58
13. Sheth PR, Tossounian J. The hydrodynamically balanced system (HBSTM): A novel drug delivery system for oral use. Dev Ind Pharm. 1984; 10(2): 313-339.
14. Prajapati SK, Kumar S1, Sahu VK2, Prakash G. Proniosomal gel of flurbiprofen: formulation and evaluation. J. Drug Del. & Therapeutics; 2012; 2(1): 1-5.
15. Hemant n. Patil, sharwaree r.Hardikar, ashok v. Bhosale. Formulation development & evaluation of proniosomal gel of carvedilol Int. J. Pharm. and Pharma. Sci, 2012; 4(1): 191-197.
16. Abdelbary.G, El-gendy N. Niosome-Encapsulated Gentamicin for Ophthalmic Controlled Delivery. *AAPS PharmSciTech,* 2008, 105 8-9105.
17. Alsara IA, Bosela AA, Ahemed SM, Mahrous GM. Proniosomes as a drug carrier for trandermal delivery of ketorolac. Eur J Pharm and Bio, 2005; 59: 485-490.

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